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Autophagy machinery genes are differentially required for autophagy and Parkin-mediated mitophagy.

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Autophagy is a highly conserved process from yeast to mammals that recycles proteins and organelles in response to various stresses such as nutrition depletion, unfolded protein or protein aggregation formation and pathogen infections. ULK1 complex consisting of ULK1/ATG13/ATG101/FIP200 in mammals is the key component in the most upstream initiation pathway that triggers downstream PI3K complex activation (containing Vps34/Beclin1/ATG14/p150) through phosphorylation of Beclin1 to induce autophagy membrane initiation. Active Vps34 then generates PI(3)P that becomes a docking point to recruit other proteins, mainly the uniquitin-like conjugation system such as ATG12-ATG5 conjugates to lipidate ATG8/LC3 to complete the closure of the autophagysomes. ULK1 complex also phosphorylates the only membrane-spanning member ATG9A to assist efficient LC3 recruitment. Genetic studies with knockout mice have suggested essential roles of each autophagy machinery genes at each step. However, recently studies suggested that each pathway seems to be independently recruited to mitochondria during Parkin-mediated mitophagy. By generating ATG5, ATG9A, ATG13, ATG14 and FIP200 single, double, pentaKO in HeLa cells, we found that unlike in mouse, different ATG genes are differentially required for autophagy and mitophagy. While ATG5, ATG9A and FIP200 KO completely block p62 degradation during starvation, ATG13 and ATG14 KO exhibits mild inhibition. ATG9A, ATG13 and ATG14 KO also display mild inhibition of mitophagy, while ATG5 KO shows moderate block and FIP200 exhibit the strongest block of mitophagy, almost to the same extent as ATG5/9A/13/14 QKO. While ULK1/ULK2 both seem to be dispensable for autophagy from other studies, the differential requirement of ATG13, ULK1/2 and FIP200 for autophagy and mitophagy that are components of the same complex suggest a complex regulation of autophagy and mitophagy than previously believed.

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Esm1p, a novel regulator of phospholipid imbalance-induced microlipophagy.

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Phospholipid imbalance due to inhibition of phosphatidylcholine (PC) biosynthesis leads to disruption of endoplasmatic reticulum (ER) morphology, activation of the unfolded protein response (UPR) and accumulation of lipid droplets (LDs) in the budding yeast, Saccharomyces cerevisiae. We previously discovered that yeast cells adapt to lipid imbalance produced by inhibition of PC biosynthesis and that formation of LDs is essential for survival and adaptation to lipid imbalance. We also found that LDs produced in response to chronic lipid imbalance are degraded by a process that resembles microautophagy, which we refer to as microlipophagy. Interestingly, ER heat shock proteins and ubiquitinated proteins are enriched in LDs produced in response to chronic lipid imbalance. These findings support the model that LD formation and degradation during chronic lipid imbalance is important not just for removing excess phospholipids from the ER but also for removing unfolded proteins from the organelle. We identified Esm1p as a protein that is required for efficient degradation